A Determination of the Resistance-Susceptibility Characteristics of Douglas-Fir to the Western Spruce Budworm

FINAL REPORT

A Determination of the Resistance-Susceptibility Characteristics of Douglas-Fir to the Western Spruce Budworm

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SUMMARY

Two models predicting resistance in Douglas-fir to the western spruce budworm were developed based on foliage quality and physical characteristics of the sample trees. Foliar terpene composition and concentrations, tanning coefficients, and total, soluble, and insoluble nitrogen levels were used as measurements of foliage quality. Tree age, dbh, height, crown ratio, time of budburst, and a variety of incremental growth measurements were used to determine the overall physiological and phenological condition of the sample trees. Larval budworm were reared on sample trees and the dry weights of adults resulting from these larvae were determined. Multiple correlation analysis was performed between adult dry weights (dependent variables) and foliage quality and other tree parameters (independent variables), to determine the degree of resistance of Douglas-fir to the budworm. Female budworm from trees high in foliar concentrations of beta-pinene, myrcene and total nitrogen weighed less than those from trees lacking these characteristics. Male budworm from trees high in foliar concentrations of alpha-pinene, myrcene, terpinolene, citronellyl acetate, and bornyl acetate weighed less than those from trees lacking these characteristics. Additionally, relatively vigorous and productive trees tended to be less susceptible to budworm of either sex. From these data it appears that relatively highly productive trees possessing certain quantities of foliar terpenes are more resistant to larval budworm feeding. These data suggest that any practice that will improve tree productivity and vigor, such as selective thinning or replanting, would enchance tree resistance and adversely influence budworm growth.

I. INTRODUCTION

1.1. The Effects of Western Spruce Budworm Damage

The western spruce budworm (Choristoneura occidentalis Freeman: Tortricidae) is a severe defoliator of several members of the Pinaceae (Johnson and Denton, 1975). The principle host trees in the western U.S. are: Pseudotsuga menziesii var glauca (Beissn.) Franco, Abies concolor (Gord. and Glend.) Lindl., Abies grandis (Dougl.) Lindl., Abies lasiocarpa (Hook) Nutt., Picea engelmanii Parry and Larix occidentalis Nutt. (Johnson and Denton, 1975).

Damage to the host tree may be minimal or severe depending on the length of time the host tree is subjected to infestation, and is usually a direct result of defoliation. At low levels of infestation, damage seems to be primarily a reduction in the amount of photosynthates. As defoliation increases with increasing populations of budworm, tree reproduction, vegetative growth, and wood production are decreased. Eventually, if defoliation is maintained, roots rot, abnormal budding may develop, infestation by bark beetles may occur, and finally, the host tree may die (Johnson and Denton, 1975).

Outbreaks of western spruce budworm, especially on the above preferred host species, have been reported in the western U.S. since 1909 (Johnson and Denton, 1975). Budworm populations typically exhibit a cyclical pattern of endemic population levels growing quickly to epidemic levels followed by a crash phase, returning to endemic levels (Johnson and Denton, 1975). The majority of outbreaks last from one to five years, although outbreaks lasting up to twenty or more years are not uncommon (Johnson and Denton, 1975, Fellin, personal communication).

1.2 Concepts of Resistance

The concept of herbivore resistance seems to be understood poorly at best. According to Painter (1951), resistance "may be defined as the relative amount of heritable qualities possessed by the plant which influence the ultimate degree of damage done by the insect." Similarly, Beck (1965) defines resistance as "the collective heritable characteristics by which a plant species, race, clone or individual may reduce the probability of successful utilization of that plant as a host by an insect species, race, biotype, or individual." Using either definition, the concept to note is that characteristics which confer resistance must be heritable, and must ultimately reduce the amount of insect damage to, and use of, the host plant that would otherwise occur without such characteristics. Specifically, in this study, resistance characteristics will be considered as those which are correlated with decreased insect growth (as measured by adult weight).

Painter (1951) considers resistance to be composed of three major components: (1) tolerance by the host plant towards the insect herbivore, (2) preference and nonpreference by the insect towards the host plant, and (3) antibiosis exhibited by the host plant towards the insect. Resistance through "tolerance" is simply the ability of the host plant to grow and reproduce while supporting an infestation of insects that would otherwise damage susceptible plants. Resistance through "nonpreference" (susceptibility being "preference") is simply those "plant characteristics and insect responses that lead...away from the use of a particular plant or variety." Antibiosis as a basis of resistance refers to any adverse effects developing from feeding on a host plant which affect the feeding insect's size, mortality, or life history.

Maxwell (1972; see also Feeny, 1970) considers in the concept of resistance the nutritional quality of the host and its effect upon the

herbivore. Host plants with proper quantitative and qualitative levels of vitamins, minerals, carbohydrates, amino acids, and fats should be initially selected ("preferred") by the herbivore. On the other hand, plants lacking these essential compounds will not be selected ("nonpreferred") or, if selected, will exhibit "antibiotic" effects as defined by decreased insect size and increased insect mortality.

Maxwell (1972) also introduces the confounding influences due to the morphology and secondary chemistry of the host tissue. The shape, color, and even the texture can influence the resistance or susceptibility of a host by making it difficult to find, or digest (Cates and Alexander, 1982; Feeny, 1970; Hanover, 1975; Kasting and McGinnis, 1959, 1961; Kasting et al., 1958; Painter, 1951; Rhoades and Cates, 1976; Southwood, 1973). Most herbivores, however, are finely tuned to their host's morphology. They have developed specific search images through the use of approproate thigmotropic, chemotropic, and phototropic responses for detecting and locating food sources (Schoonhoven, 1973).

The effects of secondary chemistry on the foliage quality of the host plant and ultimately the insect herbivore are poorly understood. It is known that certain secondary chemicals will enhance susceptibility (i.e., insect preference) of the host. These substances seem to act as cues or feeding stimulants for the herbivore (Rhoades, 1979; Rhoades and Cates, 1976; Schoonhoven, 1973). For example, mustard oil glycosides seem to enhance feeding by crucifer feeders (Janzen, 1979; Nayar and Thorsteinson, 1963). The spotted cucumber beetle's feeding is enhanced by cucurbitacins (Chambliss and Jones, 1966), and the cotton boll weevil's feeding is enhanced by gossypol (Maxwell et al., 1963). Obviously the above are examples of Painter's preference component to resistance.

Previous research in the area of foliage quality and its associated effects on forest herbivores has resulted in some promising findings. Studies conducted on Picea rubrens and Picea mariana indicate that the toxic glycoside, pungenin, is associated with resistance to the spruce budworm (Heron, 1965). Heron (1965) found pungenin when incorporated in an artificial diet to be a deterrent to budworm. Wilkinson (in Hanover, 1975) found a chemical, similar to pungenin, which exists in relatively high concentrations in the resistant P. mariana. The same compound was found in low concentrations in the susceptible P. rubrens. Albert and Jerrett (1981) found Choristonuera fumiferana to prefer the water soluble extracts from the foliage of various conifers. Work dealing with the hickory--elm bark beetle interaction suggests that resistance is conferred through the presence of juglone, a 1,4-napthoquinone (Norris, 1972). Feeny (1970) has shown that larvae of the winter month, Operophtera brumata do not feed on the mature foliage of oak due, in part, to the high tannin concentration found in the foliage. Cates et al. (1982a) found that western spruce budworm weights were positively associated with increasing foliar levels of alpha-pinene and soluble nitrogen while being inversely associated with increasing levels of beta-pinene, bornyl acetate, and other terpenes.

In order to demonstrate true resistance, however, these plant defensive mechanisms must be controlled genetically (Beck, 1965; Cates and Alexander, 1982). That these characteristics are gentically controlled is reasonably well established. Also well established is the fact that the abiotic environment, as well as biotic influences, certainly have the ability to modify the genetic influence (Cates et al., 1982a). Painter (1966) suggests demonstrating heritability by determining that the components of resistance

are present in different environments. This has already been established for Douglas-fir through stress experiments (Cates et al., 1982a) and geographically distinct chemical patterns found among populations of Douglas-fir (Von Rudloff, 1972, 1973a and b).

1.3 Objective

The major objective of this investigation was directed toward determining if the western spruce budworm is affected by the foliage quality characteristics of Douglas-fir trees. Determination of the effects of variartion in nutritional and resistance-susceptibility characteristics (i.e., the foliage quality characteristics as defined by the nitrogen, monoterpene, and resin acid content, as well as the foliar protein complexing capacity) found in the foliage of Douglas-fir trees upon western spruce budworm larval growth and adult fecundity was emphasized.

This objective was accomplished by investigating the following questions:

1. Within a given population of Douglas-fir trees, were some members of the population relatively reistant to budworm damage?
2. Does resistance to budworm in Douglas-fir result in reduced growth and fecundity of budworm feeding on such trees?
3. If some members of a population of Douglas-fir were resistant to budworm, what were the components of this resistance?

Questions 1 and 3 were tested through the use of multiple correlation techniques. Specifically, the null hypothesis that budworm fitness is not correlated with a variety of possible resistance characteristics was tested. Question 2 was tested through the use of budworm fecundity studies and simple linear regression techniques.

II. MATERIALS AND METHODS

2.1 Site and Tree Selection

The research site selected is located within Barley Canyon, in the Santa Fe National Forest, approximately 2.4 km northeast of Fenton Lake, New Mexico. The site lies along the valley floor and is approximately 3.2 km long and 0.5 km wide (1.6 km sq.). The average elevation of the site is 2440 m. The doninant tree species include Pseudotsuga menziesii var. glauca (Beisn.) Franco, Abies concolor, (Gord. and Glend.) Lindl. and Populus tremuloides Michx. At lower elevations (2375 m) Pinus ponderosa Dougl. exists while Picea pungens Englem. appears at higher elevations (2500 m) within the site. The research site was selected such that there was a history of extremely low levels of budworm defoliation. This was verified by Federal Insect and Disease Management personnel, Region 3, Albuquerque, New Mexico. Budworm were present within the site but at very low levels.

Initially, 200 Douglas-fir trees of visually similar height, dbh, and crown diameter were chosen. Variability among trees was reduced by choosing only those trees with similar microenvironments (proximity to drainage, angle and attitude of slope, and distance to neighboring trees). Following the visual selections, the actual physical parameters of height, age, and dbh were determined using clinometers, increments borers, and dbh tapes. In order to accurately determine the effects of foliage quality on spruce budworm, it was critical to further minimize the variation between trees while maximizing sample number. This was done by removing from the analyses those trees whose physical paramters deviated most among the original 200 trees. Only 105 trees met these criteria of uniformity.

2.2 Physical and Phenological Tree Measurements

In addition to age, height, and dbh, the live crown diameter, tree bole radius as calculated from the increment cores, five year growth increment (the combined width of the last five years' growth of annual rings) and crown ratio (crown height divided by tree height) were determined for each sample tree. Additionally, after foliage growth had ceased in early August, 1980, the average internodal growth for 1979 and 1980 were determined. These variables were calculated by taking the mean of three internode length measurements for each year; 1980 being the latest internode and 1979 being the internode immediately adaxial to the 1980 internode. The average internode length variables were used as estimates of the amount of tissue available to budworm in 1979 and 1980. Finally, the time at which each sample tree began to flush new tissue was noted as days since June 1, 1980.

2.3 Insect Selection and Placement

Budworm larvae to be used in the study were collected June 22, 1980 from a heavily infested area within the Carson National Forest near the Palo Flechado pass (approximately 32.2 km east of Taos, New Mexico). Branches from the heavily infested trees were cut using pole cutters, placed in insulated, air conditioned trucks, and then rushed to an environmental chamber (22°C, 35% r.h.) housed in the Biology Department, University of New Mexico. Transport was done during the cooler, early evening hours to minimize larval stress. On June 23, within 12 hours of collection and transport, larvae were removed from the infested branches and sorted by instar. Approximately 3000 fourth instar larvae were collected and placed into plastic vials containing a 5 cm length of young Douglas-fir foliage. Vials were capped with perforated plastic lids to prevent larval escape yet still provide oxygen for the insects. No attempt

was made to sort larvae by sex, and it was assumed equal numbers of both sexes were collected. Twenty-five larvae were placed into each vial. Vials were then returned to the environmental chamber to minimize larval stress.

On June 24, the experimental animals were transported to the Barley Canyon site and placed on the experimental trees. Five larvae were placed in a nylon screen bag enclosing one branch containing at least 10 foliage buds. Any local endemic larvae present on the branches were removed prior to bagging. Five bags with five larvae each were placed on each tree yielding a total of 25 larvae placed per tree. All bags were placed within the same north facing quarter of each tree at mid-crown level. The nylon bags used, while preventing larval escape, allowed 70% of full sunlight to pass through them. Since maximum photosynthesis can occur at levels as low as 20% of full sunlight in C_3 plants (Noggle and Fritz, 1976), effects due to a possible reduction in photosynthesis due to bagging were not expected.

2.4 Insect Measurements

Experimental larvae were allowed to feed on sample trees until pupation. All larvae pupated within three weeks of the time they were placed on the trees. On 11 July 1980 the bags, now containing pupae, were cut from the trees using pole cutters and transported back to the laboratory. Pupae were removed from the bags and placed in labeled paper cups and sealed with cheesecloth to prevent escape of emerging adults. Cups were placed in an environmental chamber (22°C; 35% r.h.), and adults were allowed to emerge. Animal survivorship and mortality were determined per tree based on the original 25 animals placed on each tree.

Approximately 95% of the adults that emerged, did so within 72 hours of one another indicating similar developmental rates among budworm (i.e., the

developing larvae and pupae were subjected to similar developmental times and temperatures). Differential development due to differential accumulation of degree-days was not expected.

Upon emergence, all adults were killed using ethyl acetate killing jars. Adults were sexed, weighed, and assigned identification numbers. All males were placed immediately into 10 ml glass vials and dried for 72 hours at 60°C so that dry weights could be determined. The subsample of 81 females was used for budworm fecundity determinations (see below) prior to dry weight determination.

2.5 Determination of Budworm Fecundity

For the female adult budworm choosen to determine fecundity, internal egg masses from each female were carefully dissected, removed, placed into small preweighed aluminum weighing boats, and dried for 72 hours at 60°C. Internal egg mass dry weight was then determined. The remaining portion of the female also was dried at 60°C for 72 hours so that dry weights could be determined. Internal egg mass dry weight plus the remaining portion of the female were summed to yield total female dry weight. For 30 of the original 81 females choosen, the individual eggs per internal egg mass were counted prior to drying. Therefore, for 81 females, adult dry weight and internal egg mass dry weight were determined, and for 30 females, female dry weight, internal egg mass dry weight, and numbers of eggs per internal egg mass (number of eggs per female) were determined.

2.6 Foliage Collection

During the later instars (fifth and sixth) when feeding is greatest, appproximately 30 g fresh weight of new (current year's) foliage was collected

from each tree. All foliage was collected on the same side and midcrown level of the trees where the experimental insects were placed. Collected tissue was placed in labeled zip-lock plastic bags, put on ice, taken to the laboratory and frozen. Additionally, one six inch spray bearing current year's foliage was clipped from each sample tree for subsequent monterpene analysis. From this spray, 30 milligrams of young foliage was weighed to the nearest 0.1 mg using a Cahn electric millibalance. The weighed tissue was then encapsulated within a section of indium tubing (1.0 inch x 0.120 inch o.d. x 0.080 inch i.d.). Encapsulated samples were placed in individual labeled vials and kept on ice until they could be returned to the laboratory and frozen (-20°C). To prevent breakdown or loss of monoterpenes, tissue collection, weighing, and encapsulation for monoterpene analysis were all conducted in the field.

2.7 Chemical Methods

2.7.1 Total and Soluble Nitrogen Analysis

Approximately 5 g of the current year's foliage was weighed to the nearest 1.0 mg and dried at 60°C for 72 hours. Dry weight and percent water content determinations were then made. The dried foliage was ground to a powder using a Pitchford ball mill and split into two approximately equal subsamples. Each subsample was weighed to the nearest 1.0 mg. The first subsample was used to determine total nitrogen content. This was carried out by standard microkjeldahl digestion of the dried plant material with subsequent analysis for nitrogen using a Technicon autoanalyzer. The second subsample was used to determine the insoluble nitrogen content of the foliage. The dried tissue was extracted 3 times (each extraction for 24 hours) with 95% ethanol to remove the soluble nitrogen fraction from the tissue. The first extraction used boiling ethanol, while the latter two used ethanol at room

temperature. After the last extraction, the tissue was again dried at 60°C and reweighed. Insoluble nitrogen was determined from this tissue using microkjeldahl digestion and subsequent autoanalysis. The soluble nitrogen fraction was determined by calculating the difference between total and insoluble nitrogen contents.

2.7.2 Resin Acid Analysis

Approximately 10 g samples of the current year's foliage were used for resin acid determinations. Samples were shipped to and analyzed in the laboratory of Dr. D.F. Zinkel, Forest Products Laboratory, USDA Forest Service, Madison, Wisconsin. The resin acid content of these samples was negligible, and consequently, further investigations with resin acids were not pursued.

2.7.3 Protein Complexing Compound Analysis

The method used to determine the quantity of protein complexing compounds ("tannins") was a modification of the method used by Bate-Smith (1973). In this method human hemoglobin is used as a chromoprotein to determine the quantity of protein complexing substances in an extract via colorimetry. Condensed tannins, known to be in Douglas-fir, and a variety of other hydroxyl bearing compounds, will complex with hemoglobin and precipitate from solution, resulting in a detectable color change of the solution. The more protein complexing compounds in solution, the greater the intensity with hemoglobin, and the clearer the solution becomes. Using standard curves, the amount of color change can be directly related to the quantity of tanning compounds in solution.

2.7.3.1 Blood Preparation

Ten milliliters of blood are drawn and immediately diluted to 500.0 ml with cold (4°C) distilled water. This suspension was then centrifuged for 30 min at 5000 rpm (4080Xg) to remove any particulate matter. After centrifugation, the absorbance (at 578 nm) of the blood solution was adjusted by the addition of distilled water such that, when mixed in a ratio of 3:3:1 (adjusted blood solution:distilled water:buffer solution; see reaction mixture described in "Generation of Standard Curve" below) yielded an absorbance of 1.00. This adjusted blood solution can be used for at least five days before significant deterioration occurs (Redak, unpublished data).

2.7.3.2 Generation of Standard Curve

Standard solutions of tannic acid were prepared at the following concentrations: 0.0, 10.0, 20.0, 30.0, 40.0, 50.0, 60.0, 70.0, 80.0, 90.0, and 100.0 mg tannic acid per 100.0 ml distilled water. Three milliliters of adjusted blood solution, 3.00 ml of a given tannic acid standard solution, and 1.00 ml of 0.1N citric acid-sodium citrate buffer (pH=5.5) were combined. This mixture was mixed vigorously in a 15 ml test tube for 30 seconds using a vortex mixer and then centrifuged for 20 minutes at 3000 rpm (1085Xg). Protein complexation and precipitation occurs during this period. After centrifugation, the supernatant was collected, and its absorbance read at 578 nm. A standard curve was generated by running triplicate determinations on each of the above 12 tannic acid solutions and plotting the absorbance values obtained versus the tannic acid concentrations of the solution (Figure 1). The blood solution was adjusted such that the absorbance values for the tannic acid solution containing 0.00 mg tannic acid per 100.0 ml of distilled water

were 1.00. A new standard curve must be prepared with each new batch of blood solution.

2.7.3.3 Determnation of Protein Binding Compounds

Approximately 5.0 g of the current year's tissue was weighed to the nearest 1.0 mg and frozen in an ultra-cold freezer (-80°C). Due to the extremely low temperatures, the frozen tissue was ground easily to a powder using a precooled (-80°C) mortar and pestle. Ground tissue was quantitatively transferred to a 250 ml Ehrlenmeyer flask. One hundred milliliters of a 50:50 methanol to water solution were added to the sample flask. The flask was placed in a 55°C shaker bath for 90 min during which time the protein complexing compounds were allowed to diffuse into the extraction solution. To prevent any oxidation of the extracted compounds, nitrogen was passed over the sample for the entire extraction period.

After the 90 min extraction, the sample was removed from the shaker bath and filtered using Whatman number 1 filter paper. The extract solution was then flash evaporated to approximately 2.0 ml. The sample was brought to a 10.00 ml volume using distilled water and transferred to a nalgene centrifuge tube. The 10.0 ml extract was centrifuged for 30 minutes at 12,000 rpm (17,300Xg) to remove any remaining particulate matter. The supernatant was collected and again brought quantitatively to a 10.0 ml volume. In order for the sample extract's absorbance to fall within the linear portion of the standard curve 1.00 ml of this final extract was diluted with 4 ml distilled water. Protein complexing capacity was then determined using this final diluted sample.

A reaction mixture, containing 3.00 ml of adjusted blood solution, 1.00 ml of citric acid-sodium citrate buffer (pH=5.5), and 3.00 ml of the diluted

sample extract was mixed for 10 seconds using a vortex mixer and centrifuged for 20 min at 3000 rpm (1085Xg). The supernatant was carefully collected and its absorbance read at 578 nm. Sample absorbance was converted to tannic acid milligram equivalents per 100 ml using the standard curve. After adjusting for dilutions and sample weights, results were reported using milligram equivalents of tannic acid per gram fresh weight of tissue.

2.7.4 Monoterpene Analysis

Samples were prepared for gas chromatographic analysis by removing collected tissue from the indium tubing capsules and reweighing to the nearest 0.1 mg. Solid sampling and column loading were performed by initially placing the sample tissue into a glass gas chromatography injector. Then, for a period of 10 minutes, monoterpenes from the tissue were loaded via volatilization onto the column using a helium flow rate of 1.0 ml/min and an injector temperature of 175°C. During this loading period the column oven was maintained at 35°C to prevent movement of monoterpenes through the column.

After the loading period the helium flow rate was reduced to 0.0 ml/min, and the now dried tissue removed from the injector and weighed. Monoterpene analysis was allowed to continue using a Perkin-Elmer Sigma 2B gas chromatograph equipped with flame ionization detectors linked to a Perkin-Elmer Sigma 10B Data Interpretation Station. The column was a Perkin-Elmer 0.010 inch by 150 foot capillary column packed with 85% OS-138, 14% Co-880, and 1% V-930. Helium flow for the analysis (following loading) was brought back up to 0.5 ml per minute. Temperature programming was as follows: Initial temperature (after loading), was set to 45°C. This was followed by an immediate increase of 0.5°C per min to 55°C, and another immediate increase of 2.0°C per min to 150°C, then 20.0 min at 150°C.

Standards were used to identify alpha-pinene, beta-pinene, myrcene, camphene, beta-phellandrene, 3-carene, terpinolene, gamma-terpinene, limonene, bornyl acetate, citronellyl acetate, and geranyl acetate. Compounds which were unknown were assigned an identification number according to their retention times. The concentration of each compound was determined as area counts. These counts subsquently were standardized, relative to dry weights, and results were reported as counts per 20 mg dry weight of tissue.

2.8 Budworm Fecundity Measurements

Simple linear regressions were performed to determine the relationships between adult female budworm dry weight, weight of total egg mass within each female, and the total number of eggs within each female. These analyses were performed to determine if adult female dry weight was a valid estimator of adult fecundity. In this study, fecundity is assumed to be closely and positively correlated with fitness.

Initially to test the null hypothesis that there was no relationship between total egg mass dry weight and adult female dry weight, total internal egg mass dry weight was used as the dependent variable and regressed upon adult female dry weight (n=81). It was expected that larger females produced larger internal egg masses.

Secondly, to test the null hypothesis that there was no relationship between total number of eggs per female (which equals the total number of eggs per internal egg mass) and total internal egg mass dry weight, the number of eggs dissected from each female was used as the dependent variable and regressed upon total internal egg mass dry weight (n=30). If internal egg mass dry weight was positively associated with female dry weight, then it was critical to determine if increased egg mass dry weight was due to an increase

in the total number of eggs per egg mass or an increase in the size of eggs per egg mass.

To test the null hypothesis that there was no relationship between the total number of eggs per female and adult female dry weight, the number of eggs dissected from each female was again used as the dependent variable and regressed upon adult female dry weight (n=30). If the two preceding simple regressions were both significant with a positive slope, then the relationship between adult female dry weight and the number of eggs per female also should be positive. Larger females should produce larger egg masses consisting of a higher number of eggs than should smaller females. If the above is true, then larger females should be more fecund (fit) than smaller females. It is assumed that fecundity is a valid estimator of fitness. However, McKnight (1971) has suggested that fitness may be lower in highly fecund females in heavily defoliated forests due to excessive mortality of subsequent offspring. Since Barley Canyon had not experienced severe defoliation, it was felt this was not a cause for concern.

2.8 Multiple Correlation Analyses

Measurements of the 34 parameters (Table 1) were used as independent variables in a multiple stepwise correlation analysis. In all, 23 variables were used to determine foliage quality characteristics, and 11 variables defined the physical and phenological characteristics of the sample trees.

Two correlation analyses were performed using two different dependent variables. The dependent variables used in the analyses were the average adult female budworm dry weight and the average adult male budworm dry weight. These dependent variables were determined by calculating the average female

and male adult budworm dry weights per sample tree based on those adults which were raised as larvae on the sample trees.

All analyses were performed using either BMDP or SAS statistical programs on a IBM 3032 mainframe computer.

Summary statistics for all variables measured may be found in Appendix I.

III. RESULTS

3.1 Budworm Fecundity Measurements

The regression analyses used to determine female budworm fecundity indicated that total internal egg mass dry weight was correlated positively with female dry weight (Figure 2). Additionally, the number of eggs per female (which equals eggs per internal egg mass) was correlated positively with the total internal egg mass dry weight (Figure 3), and the total number of eggs per female also was correlated positively with female dry weight (Figure 4). The results of these analyses indicate that heavier females contain a heavier internal egg mass, and that heavier egg masses consist of more eggs rather than heavier eggs (Figure 2 and 3). Hence, heavier females contain a larger number of eggs per female than lighter females (Figure 5).

3.2 Multiple Correlation Analyses

Of the 34 independent variables that were initially thought to be important in explaining adult female budworm dry weight, 11 variables explained 34.9% of the total variation (Table 2). Again, bornyl acetate, unknown terpene number 5, and total foliar nitrogen content were the significant variables in this model (as determined by t-test; Zar, 1974). Average adult female dry weight was associated positively with age, average internode length for 1980, time of budburst, terpinolene, bornyl acetate, and geranyl acetate. Five year growth increment, total foliar nitrogen content, beta-pinene, myrcene, and unknown terpene number 5 were correlated negatively with average adult female dry weight.

Examination of the standardized correlation coefficients for the model for female dry weight showed bornyl acetate, unknown terpene number 5, total nitrogen content, and beta-pinene as being the most important variables in

predictng the dependent variable (Table 2). Time of budburst, terpinolene and myrcene were of intermediate importance, while age, geranyl acetate, five year growth increment, and average internodal length for 1980 were of relatively minor importance in predicting average adult female dry weight.

Of the 34 independent variables initially used to predict average adult male dry weight, 15 variables explained 35.5% of the total variance in the model (Table 3). Here, limonene, terpinolene, and citronellyl acetate were significant variables within the model (as determined by t-test, Zar, 1974). Average adult male dry weight, was associated positively with average internode length for 1980, percent water content of the current year's foliage, the ratio of soluble to insoluble foliar nitrogen, limonene, gamma-terpinene, and unknown terpene numbers 4, 5, and 9. Crown ratio, alpha-pinene, mycrene, terpinolene, bornyl acetate, citronellyl acetate, and unknown terpene number 8 were related inversely to adult male dry weight.

Examination of the standardized correlation coefficients for the male model (Table 3) showed that limonene, terpinolene, citronellyl acetate, alpha-pinene, and bornyl acetate were of major importance in predicting adult male dry weight. Myrcene, percent water of the current foliage, gamma-terpinene, unknown terpene numbers 8 and 5 were of moderate importance, while crown ratio, unknown terpene numbers 4 and 9, the ratio of soluble to insoluble foliar nitrogen levels, and the average internode length for 1980 were of relatively little importance in predicting adult male dry weight.

IV. DISCUSSION

Examination of Figures 2 and 4 shows that female dry weight is a valid estimator of fecundity. It is assumed, that in endemic or expanding budworm populations that fecundity is related positively to female fitness. Fitness in this study will be defined as the genetic contribution of an individual to future generations (Ricklefs, 1973). That is, the more fecund a female budworm is, the more fit she is. This may not be the case with epidemic budworm populations in already heavily defoliated forests. McKnight (1971) has found that the survivorship of offspring from such epidemic populations is considerably lower than those of low density expanding populations. While fecundity of the endemic and epidemic population types seems to remain the same, survivorship is lower in the epidemic population. Hence, fitness is reduced in individuals of the epidemic population. The research site used in this study showed little sign of budworm infestation during 1980. Therefore, the assumption that increased fecundity (as indicated by increased female dry weight) leads directly to increased female fitness seems valid for this particular study. Whether or not male budworm dry weight is indicative of male fitness is not known. Increased adult male weight may or may not lead to increased fitness depending on the species in question. For the lepidopteran species, Colias philodice and C. eurytheme, increased male weight is correlated with increased male fitness (Marshall, 1982). For the western spruce budworm, however, no known definitive study has been made to determine the relationship between male weight and male fitness. For this study, all that may be said with regard to male weight is that it is correlated significantly with numerous Douglas-fir chemical and physical measurements.

It cannot be assumed, at this point, that these possible relationships with the host tree are affecting male budworm fitness.

Examination of Table 2 which displays the model for average adult female dry weight, indicates that sample trees with high foliar levels of total nitrogen, beta-pinene, myrcene, and unknown terpene number 5, as well as a large five year growth increment may be more resistant to female budworm damage than trees without these characteristics. On the other hand, sample trees which are relatively old, flush a relatively large amount of tissue late in the season, and contain high foliar levels of bornyl acetate and terpinolene, seem to be more susceptible to female budworm damage.

That increased levels of total nitrogen may confer resistance in Douglas-fir is rather surprising. There are numerous examples in the literature suggesting that increased nitrogen levels render host plant species more palatable and nutritious, hence more suceptible to insect herbivore damage (Cates et al., 1982a; Mattson, 1980; White, 1969, 1974). One possible explanation for this result is that total nitrogen levels in nonstressed host trees may be indicative of the trees' productivity and vigor (Gosz, 1981; Mattson, 1980; Miller et al., 1969; Van den Driessche and Dangerfield, 1975). If the assumption is made that tree productivity and growth are equated with overall vigor and health, then increased foliar nitrogen levels reflect the health and vigor of the tree. Trees with high nitrogen levels are relatively healthy and vigorous and hence resistant to budworm. Trees with low levels of nitrogen should be less vigorous and more suceptible to insect attack and damage.

The hypothesis that tree vigor (as indicated by high total nitrogen levels) may be responsible for increased budworm resistance in Douglas-fir is

further supported by the fact that the five year growth increment is also correlated negatively with female budworm success. Highly productive and vigorous trees should have wider annual rings than trees which are less productive. Trees which are relatively productive have a larger five year growth increment and seem to be more resistant to female budworm damage than relatively unproductive trees with a smaller growth increment.

Tree age being positively correlated with female budworm success is not surprising. Mott (1963) points out that in the eastern sections of the United States and Canada, mature and overmature stands of balsam fir are much more susceptible to attack by the eastern spruce budworm (C. fumiferana). Mott attributes this susceptibility of older trees to the fact that such treees are usually taller with open and exposed upper canopies. This condition is thought to lead to high evaporation rates in the upper canopies, a condition favorable to the late larval instars. If that were the case in this study, one would expect tree height also be be an important variable in the multiple correlation equation, which it was not. Additionally, some authors (Mattson, 1980; Miller et al., 1979) feel that with age total nitrogen levels may become reduced leading to a decrease in productivity and vigor. If this is the case, and the assumption is made that female budworm prefer trees with lower productivity as is supported by the relationships shown for total nitrogen and the five year growth increment, then indeed, older trees should be more susceptible to female budworm damage.

The average internode length for 1980 was used as an indicator of the amount of foliage available to the experimental animals for food. The assumption was made that longer internodes should develop equal numbers of needles per linear centimeter of internode. Therefore, longer internodes will

have more needles (i.e., more available foliage for budworm utilization) than short internodes. When viewed in these terms, it should be expected that increased internode length (increased available foliage) will be correlated positively with budworm success as is the case here.

The positive relationship between time of budburst and female budworm success is in direct contradiction with earlier studies. In this study, it appears that the later a tree bursts bud the more susceptible it is to damage. For the eastern spruce budworm, however, Blais (1957) found that one of the major characteristics of budworm resistance in black spruce (Picea mariana) was that this species bursts bud late in the season. His explanation for late budburst conferring resistance was that at the time of budburst, budworm were in the later larval feeding stages, and trees which burst foliage late did not provide an adequate amount of preferred young tissue. In other words, host plants were escaping in time from their herbivore--one alternative predicted by theory (Feeny, 1976; Rhoades and Cates, 1976). Budworm growing on black spruce consistently exhibited a lower rate of development and a higher rate of mortality than those growing on the early bursting Picea alba or Abies balsamea, the preferred hosts.

In this study, it should be noted that experimental insects in the fourth instar were not placed on sample trees until all of the sample trees had burst bud. Initially, there should have been no shortage of young tissue for the experimental animals. The relationship between time of budburst and the dependent variable can be simply explained. While all animals had an adequate supply of foliage for growth, the phenological stage of the tissue eaten varied from tree to tree. According to recent theories of plant defensive chemistry (Feeny, 1976; Rhoades, 1979; Rhoades and Cates, 1976), by the time

placement of experimental animals occurred, one would expect tissue which had burst early to have developed (albeit poorly) a certain amount of defensive chemistry and structure. On the other hand, tissue which had burst late, immediately prior to the placement of animals, should have little if any herbivore defensive capability. In other words, undefended tissues were not allowed to escape in time. This is exactly what these results indicate. Females placed on older (although still the current year's) tissue exhibited lower adult dry weights than did females placed on younger (later bursting) tissue.

The relationship between beta-pinene, myrcene and unknown terpene number 5 with average adult female dry weight suggests that these three chemicals may be conferring a degree of resistance against female budworm. To some extent, this is in agreement with other studies. Cates et al. (1982a) showed that when budworm were reared on nonstressed Douglas-fir trees, adult male and female dry weights were lower as compared to animals reared on stressed (i.e., trenched) trees. Additionally, the authors found that nonstressed trees contained significantly higher concentrations of beta-pinene, bornyl acetate, and two unknown terpenes. They suggest that the higher concentrations of the reported terpenes are responsible for conferring the apparent resistance in nonstressed trees.

For this study, however, bornyl acetate, geranyl acetate, and terpinolene may confer a certain degree of susceptibility to budworm. To some extent, these results, especially those for bornyl acetate, are in contrast with the results reported by Cates et al. (1982a). One possible explanation for this deals with the geographical variation among Douglas-fir monoterpene chemistry. Von Rudloff (1972, 1973a, 1973b) has shown extensive terpene variation among

Douglas-fir populations over a matter of only a few kilometers. Additionally, current plant herbivore theories and studies suggest that local adaptation by herbivores to their host plant(s) does occur (Cates and Rhoades, 1977; Feeny, 1976; Rhoades and Cates, 1976). Therefore, given populational terpene diversity and local herbivore adaptation to these terpenes, it is possible for an individual terpene to confer both resistance and susceptibility within a host species. Whether or not a specific terpene confers resistance or susceptibility within a population, however, should be fixed to some degree. The study performed in Barley Canyon and the study performed by Cates et al. (1982a) although geographically in the same forest district, were probably from two different populations of trees. Therefore, differences in herbivore response between these populations of trees is to be expected. Similar patterns in population divergence of terpenes of ponderosa pine was suggested by Sturgeon (1979) with regard to bark beetle attacks.

Examination of Table 3, the model for average adult male dry weight, indicates that sample trees with high foliar levels of alpha-pinene, myrcene, terpinolene, bornyl acetate, citronyl acetate and unknown terpene number 8, as well as a large crown ratio, may be more resistant to male budworm damage than trees that are lower qualitatively in these characteristics. Additionally, sample trees with high foliar levels of limonene, gamma-terpinene, unknown terpene numbers 4, 5, and 9, as well as a high foliar water concentration, a high soluble to insoluble foliar nitrogen ratio, and a relatively long 1980 internode length seem to be more susceptible to male budworm.

To a certain degree, the results for the male budworm model agree with the results obtained for the female model. While total foliar nitrogen content did not enter into the male model, the ratio of soluble to insoluble

(i.e., structural) foliar nitrogen concentrations did enter. Although, of only minor importance, the ratio of soluble to insoluble foliar nitrogen did show a positive correlation with respect to average adult male dry weight. This is in agreement with other studies which suggest that as a plant's soluble nitrogen levels increase and/or insoluble nitrogen levels decrease, the susceptibility to herbivore damage will increase (Cates et al., 1982a; Mattson, 1980).

Many studies have indicated that the behavior of nitrogen levels within a plant is related to drought stress (Saunier et al., 1968; Van den Driessche and Weber, 1975; White, 1969, 1974). That is, as stress increases, soluble nitrogen levels also will increase while insoluble nitrogen levels decrease. Stressed plants should then become more susceptible to herbivore damage. Assuming that the productivity of a stressed plant is lower than a nonstressed plant, one would expect, based partially on the results of the female budworm model, that herbivore damage would be greater on the less productive stressed plants. Indeed, the male budworm model supports this view. The variable crown ratio, which is the height of the live crown of the tree divided by the tree height, entered into the male model with a negative correlation coefficient. If it can be assumed that healthy, vigorously growing productive trees have larger crown ratios than poorly growing, unproductive and possibly stressed trees, productivity should be correlated inversely with male budworm success. Also, it should be noted that there is an inverse relationship between crown ratio (a measurement of productivity) and the ratio of soluble to insoluble nitrogen (an indirect measurement of stress) as indicated by their partial correlation coefficient (partial correlation coefficient=0.202, p.=0.0674). While not significant at the 0.05 level, this relationship still

suggests that as stress increases, productivity decreases. This information coupled with the results thus far derived from the multiple correlations indicates that stressed, low productive trees are more susceptible to budworm damage.

As in the female model, the average internode length for 1980 was associated positively with the dependent variable, average adult male dry weight. Again, the assumption is made that average internode growth for 1980 was used as an indicator of the amount of foliage available to the experimental animals for food. It is not surprising, then, that increased internode length (i.e., increased available food) leads to larger males feeding on such trees.

The positive relationship exhibited by the foliar water content (% fresh weight) with average adult male budworm dry weight is not unusual. Feeny (1970) showed that winter moth caterpillars (Operophtera brumata L.) fed on and preferred the younger foliage of oak. This preference was attributed to the fact that as oak foliage matured, it exhibited increasing levels of polyphenolics, increasing tissue toughness, and decreasing water content. That is, winter moth larvae preferred only tissue which was tender, low in polyphenolics and high in water content. Male budworm, while not showing any kind of relationship with polyphenolics, did show a positive relationship with water content. Tissue toughness was not measured and was assumed to be inversely correlated with water content (Feeny, 1970).

The relationship between alpha-pinene, myrcene, terpinolene, bornyl acetate, citronellyl acetate and unknown terpene number 8 with adult male average dry weight suggests that these chemicals may be conferring to Douglas-fir a certain degree of resistance against male budworm. To a large

extent, this contrasts with the results for the above female model where bornyl acetate and terpinolene were positively correlated with increased susceptibility. However, the male budworm model is in agreement with the female model in that both models suggest myrcene may confer resistance. The inverse relationship exhibited by alpha-pinene with average adult weight is in contrast with the study performed by Cates et al. (1982a). These authors did, however, suggest an inverse relationship between bornyl acetate and budworm success as was the case in the male correlation model presented here.

For the male budworm model, limonene, gamma-terpinene, and unknown terpene numbers 4, 5, and 9 are correlated positively with the dependent variable indicating that trees with high levels of these compounds are relatively susceptible to damage by male budworm. These relationships only slightly contrast with those found for the female model. Here unknown terpene number 5 is thought to confer susceptibility against male budworm, while in the female model, terpene number 5 confers resistance to female budworm. The other terpenes conferring possible resistance to male budworm did not enter into the female model above or the model put forward by Cates et al. (1982a).

Differences between the results of the male adult budworm model and the results obtained by Cates et al (1982a) may be explained (as were the differences between the female budworm model and the results of Cates et al. (1982a) through possible differential herbivore responses to host foliage quality. This differential herbivore response may be due to local adaptation of the herbivore to differences in host plant foliage quality brought about by the natural geographical variation in terpenes among Douglas-fir populations.

Differences between the male budworm model and female budworm model are more difficult to explain. One explanation is that these differences in

results are attributable to different foliage quality requirements of the sexes. Sexual differences in feeding requirements and behaviors have been reported for species of mosquitoes and tabanids (Chapman, 1971). Unfortunately, whether or not there are sexual differences in feeding requirements for Choristoneura spp., or any tortricids for that matter, is not known. Further studies into the feeding and nutritional requirements of budworm are needed.

It is important to note that while the simplified male and female models do differ somewhat from one another, both models exhibited similar trends with regard to the independent variables measured. The four most important variables (bornyl acetate, beta-pinene, unknown terpene number 5 and the total foliar nitrogen content) are all concerned with the chemical make up of the foliage. Tree physical and phenological data (age, time of budburst, etc.), are less important in predicting female weight. Similarly for the male model, the ten most important variables in predicting the average male adult dry weight (alpha-pinene, myrcene, limonene, gamma-terpinene, terpinolene, bornyl acetate, citronellyl acetate, unknown terpene numbers 5 and 8, and the percent water content of the tissue) all deal specifically with the foliage quality of the host trees. In both cases, this is not to say the physical and phenological variables are not important, but simply, the chemical variables are relatively more important in predicting the dependent variables involved.

V. CONCLUSIONS

Within the last several years most studies investigating the interactions between the western spruce budworm and its various hosts have dealt with the causes of outbreaks and declines of epidemic budworm populations. The majority of these studies attribute climate, tree phenology, and predator and parasite interactions with the budworm as the primary determinants of budworm release and control. This study has attempted to show that host tree foliage quality may be important in determining whether or not a given tree is inherently susceptible to budworm damage.

It was shown that relatively highly productive trees exhibiting high levels of beta-pinene, myrcene and an unknown terpene number 5 are correlated with reduced biomass production and fecundity of female budworm. At endemic budworm population levels, in which this study was done, reduced fecundity of female budworm will most certainly tend to maintain low numbers of budworm feeding on a given tree. Such trees will tend to be relatively resistant to budworm. Whether or not the same type of relationships are attributable to male budworm-Douglas-fir interactions is not known since it is not known if male fitness is dependent on adult male dry weight.

It should be emphasized that this study was conducted on sample trees which had not been subjected to massive herbivore damage. If these trees were undergoing heavy defoliation, the resistance-susceptibility characteristics suggested here may change drastically. It is not known whether or not additional, perhaps more effective, resistance characteristics are "turned on" or induced under heavy herbivory. That this is the case in fir and lodgepole pine-bark beetle interactions has already been shown (Berryman, 1972; for review see Cates and Alexander, 1982).

Finally, it is important to note, that the results of this study, with proper verification, could be used by land managers and silviculturalists attempting to prevent, rather than control, outbreaks of western spruce budworm and other defoliators. These data suggest that any practice that will improve tree productivity and vigor, such as selective thinning, would enhance tree resistance and adversely influence budworm growth. Thinning of the more susceptible tree would further enhance individual tree and stand resistance. In addition to the current practice of planting extremely high yield trees, emphasis should also be place on planting trees with a diverse and effective battery of secondary compounds for deterring, or at least, minimizing budworm damage. It has already been shown that in moderate to high population level situations, Douglas-fir trees with a very diverse terpene chemistry are more resistant to budworm than trees approaching the population average of terpene chemical diversity (Cates et al., 1982b). Given that such "resistant" trees are present, when insect outbreaks do occur, the mortality and loss of yield will be less when compared to a stand of similar trees without these foliage quality characteristics. Additionally, when tree resistance factors are combined with other integrated pest management techniques, the control of insect herbivores should be easier, cheaper and tree yields should be maximized.

RECOMMENDATIONS

Additional research is needed using agar diets to determine which of the terpenes are responsible for adversely affecting the budworm. The mechanism of action of these potential toxins also needs to be elucidated. It is important to note that these compounds may only be markers of resistance and may not be the actual compounds conferring resistance. Agar diet studies, currently underway in our laboratory, would shed light on some of these questions.

Determining the foliage quality of Douglas-fir at different geographical locations across the western U.S. and how these different foliage qualities affect the associated endemic or epidemic budworm populations would be invaluable. Questions such as how variable and effective are foliage quality characteristics from area to area could be ascertained. To a certain extent this work is already being carried out. Foliage quality analysis has already been performed on one site in Montana, a site in Idaho, and four sites in New Mexico. Additionally, a study which follows the changes that may occur in the foliage quality of Douglas-fir as an associated WSB population expands from endemic to epidemic levels is needed. The opportunity for such a study now exists at the Barley Canyon site. Such a study would elucidate if resistance mechanisms are inherent, inducible, or if such mechanisms vary with changes in the physiological status of trees and of the insect population. The implications are strong from the data gathered that foliage quality characteristics can and do affect budworm growth and fecundity, but it is not known if larval population dynamics influence foliage quality characteristics. Investigations pursuing these ideas need to be continued and refined.

Following a basic understanding of the above, several management techniques could be explored with the objective of reducing the adverse effects of budworm on host plants and stands.

WORK REMAINING ON STUDY:

The current study was completed. No additional work is anticipated.

COOPERATION AND COORDINATION:

We wish to thank those scientists of Region 3 for their help before this grant was funded and their continued advice during the selection of sites. Douglas Parker and Mike Chavez of the Federal Insect and Disease Management Program, and Dan Winner, District Ranger, Jemez Ranger District, Santa Fe National Forest, were extremely helpful. We wish to extend our thanks to Dr. D. Zinkel for his analysis of Douglas-fir for resin acids.

PROBLEMS ENCOUNTERED:

No major problems of any sort were encountered. The research site was, however, moved at the last minute from Montana to New Mexico. This move was cleared through the CANUSA research director.

MANUSCRIPTS OR REPORTS PREPARED OR PLANNED:

This report is being modified appropriately for publication in Oecologia. It will be submitted for publication in about 2-4 months with the same persons as co-authors.

Table 1. The dependent and independent variables used in the multiple correlation analyses.

Multiple Correlation Independent Variables

Tree Phenological and Physical Variables	Foliage Quality Variables
age, height, dbh, crown diameter, bole radius, five year growth increment, average internode length (for 1979 & 1980), time of bud burst	alpha-pinene,* beta-pinene, camphene, myrcene, limonene, gamma-terpinene, terpinolene, bornyl acetate, citronellyl acetate, geranyl acetate, unknown terpenes #2-#5 and #7-#10 total terpenes, terpene diversity,** total nitrogen content, ratio of soluble to insoluble nitrogen, tannin content***

Multiple Correlation Dependent Variables

for females:
average adult female
dry weight

for males:
average adult male
dry weight

* all terpenes are expressed as counts/20 mg dw
** based on Simpson's diversity index.
*** expressed as mg/100 ml tannic acid equivalents.

Table 2. Female model and associated ANOVA table.

Summary Statistics:
Multiple R_2 = 0.5905
Multiple R = 0.3487

Analysis of Variance Table:

Source	df	SS	MS	F	p.>F
Regression	11	821.554	74.687	3.31	0.0011
Residual	68	1534.373	22.564		
Total	79	2355.927			

Female Model:

Variable Entered	Correlation Coefficient	Standard Error	Standardized Correlation Coefficient
Bornyl Acetate*	0.00334	0.00122	0.354
Unknown terpene #5*	-0.04165	0.01862	-0.262
Total nitrogen	-114.75060	51.62314	-0.232
Beta-pinene	-0.00541	0.00290	-0.229
Time of bud burst	0.41324	0.22878	0.193
Terpinolene	0.04044	0.02702	0.172
Myrcene	-0.01387	0.00948	-0.165
Age	0.16913	0.12242	0.145
Geranyl acetate	0.00436	0.00375	0.137
Five year growth increment	-1.59537	1.31204	-0.133
1980 internode length	0.48272	0.37711	0.133

Intercept = 23.27587

*Significant within the correlation model (t-test, p.<.05)

Table 3. Male model and associated ANOVA table.

Summary Statistics:
Multiple R_2 = 0.5955
Multiple R = 0.3546

Analysis of Variance Table:

Source	df	SS	MS	F	p.>F
Regression	15	90.543	6.036	2.49	0.0056
Residual	68	164.787	2.423		
Total	83	255.330			

Male Model:

Variable Entered	Correlation Coefficient	Standard Error	Standardized Correlation Coefficient
Limonene*	0.00512	0.00200	0.461
Terpinolene*	-0.02296	0.00999	-0.300
Citronellyl acetate*	-0.00335	0.00144	-0.256
Alpha-pinene	-0.00079	0.00055	-0.234
Bornyl acetate	-0.00062	0.00040	-0.202
Myrcene	-0.00526	0.00315	-0.192
Tissue water content	6.74118	4.43527	0.177
Gamma-terpinene	0.00824	0.00543	0.167
Unknown terpene #8	-0.02614	0.01863	-0.159
Unknown terpene #5	0.00770	0.00598	0.149
Crown ratio	-5.52496	4.97238	-0.124
Unknown terpene #9	0.00946	0.00831	0.121
Ratio of soluble to insoluble nitrogen	0.71473	0.68189	0.119
Unknown terpene #4	0.00658	0.00686	0.104
1980 internode length	0.11979	0.12284	0.102

Intercept = 7.73277

*Significant within the correlation model (t-test, p.<0.05

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Appendix I

Independent Summary Statistics for all Variables Measured.

Variable	n	Mean	Standard Deviation	Coefficient of Variation(%)
Alpha-pinene	97	1070.16[a]	525.38[a]	49.09
Camphene	97	1519.47	688.93	45.34
Beta-pinene	97	372.18	240.61	64.65
Myrcene	97	81.66	62.39	76.40
Limonene	97	449.29	153.62	34.19
Gamma-terpinene	97	60.65	42.91	70.75
Terpinolene	97	43.98	23.43	53.27
Bornyl acetate	97	638.84	580.81	90.92
Citronellyl acetate	97	34.60	124.68	360.36
Geranyl acetate	97	83.84	169.17	201.79
Unknown terpene # 2	97	36.62	47.12	128.69
Unknown terpene # 3	97	20.38	21.27	104.34
Unknown terpene # 4	97	17.17	26.41	153.85
Unknown terpene # 5	97	29.03	33.99	117.06
Unknown terpene # 7	97	3.57	10.25	287.38
Unknown terpene # 8	97	6.36	10.32	162.31
Unknown terpene # 9	97	12.55	20.98	167.23
Unknown terpene #10	97	388.71	568.37	146.22
Total terpenes	97	4870.01	1866.69	38.33
Terpene diversity	97	0.68	0.06	8.59
Total nitro.	99	1.23%dw	0.23%dw	19.05
Soluble/ insol. nitro.	98	0.36	0.28	77.82
Foliar protein complexing compounds	100	1.71%fw	0.87%fw	50.65
Water content	97	68.33%fw	4.15%fw	6.07
dbh	105	22.83cm	3.90cm	17.10
Bole radius	105	9.15cm	1.72cm	18.78
Tree height	105	11.70m	1.73m	14.80
Tree age	105	37.86yrs	4.75yrs	12.55
Crown diam.	105	5.35m	0.91m	16.91
Crown ratio	105	0.84	0.05	5.47

For All Trees (Cont.):

Variable	n	Mean	Standard Deviation	Coefficient of Variation(%)
Five year growth incr.	105	1.65cm	0.46cm	27.98
1979 internode length	105	3.76cm	1.20cm	31.88
1980 internode length	105	3.31cm	1.41cm	42.55
Time of budburst	105	13.21days	2.68days	20.32

[a] all monoterpene concentrations are reported in counts/20 mg dw.